REMARKS

Claims 81, 82, 85, 86, 102, 124, and 144-150 are currently under consideration in the instant application. Claim 1 has been amended. Support for the amendment to the claims can be found in the specification as filed. No new matter has been added.

Oath

An oath will be filed under separate cover to address the deficiency in the oath previously filed.

Sequence Listing

A sequence listing is submitted herewith.

Application Status

The specification has been amended to update the status of the US application mentioned therein.

Withdrawal of Rejections

Applicants thank the Examiner for withdrawing the provisional double patenting rejection and the various rejections under 35 USC 102.

Rejection of Claims Under 35 USC 112, First Paragraph

The Examiner has indicated that he believes that there is no support for the single-chain TCR language in claim 81 disclosing "α and β variable chain TCR covalently linked together by a second peptide linker". Applicants respectfully point to Claims 85 as previously pending for support for this amendment.

Moreover, on page 8 reference is made to U.S. application 08/943,086. At page 30, line 22, all cited references are incorporated by reference. This application sets forth constructs and linkers and provides direct support for the claim language. Additionally, specific examples of single-chain TCRs containing Vα - $(G_4S)_4$ - Vβ - Cβ domains are provided in, for example, Figure 1a, Example 1 (page 31 lines 14-18) and Example 2 (page 32 lines 7-9). In these

examples, the $(G_4S)_4$ peptide comprises the peptide linker. Additionally, Applicants disclose a single-chain TCR linked to IL-2 molecules in Examples 5 to 11.

The Examiner also believes that there is no support for the language of claims 144, 145 and 146.

Applicants point the Examiner to, for example, page 12 lines 10-15, 17 of the specification. Specifically, this passage sets forth:

> A specific example of a TCR fusion complex fused to an effector molecule is as follows: an sc-TCR such as the p264 sc-TCR disclosed below in Examples 5 below can be produced by transfecting mammalian cells with 264 DNA vector illustrated in FIG. 1. The sc-TCR p264 protein fusion complex recognizes a processed peptide fragment from human wild-type p53 tumor suppressor protein presented in the context of human HLA antigen; HLA-2.1. ... The peptide sequence is LLGRNSFEV.

Moreover, the specification discloses effector molecules as cytokines on, for example, page 10 lines 15 to 25.

Additionally, Example 1 at page 30 lines 29-32 discloses:

> The T cell clone, 264, recognizes a peptide fragment (aa 264-272; LLGRNSFEV) of the human wild-type tumor suppresser protein p53 restricted by HLA-A2.1. The T cell receptor gene was cloned into a three domain single-chain format previously shown to produce soluble TCR and functional receptor molecules.

Example 1 additionally discloses generation of the 264 single-chain TCR gene encoding the TCR that recognizes a peptide fragment (aa 264-272; LLGRNSFEV) of the human wild-type tumor suppresser protein p53 restricted by HLA-A2.1 (page 31 lines 2 to 29). Example 4 discloses identification of the 264 single-chain TCR construct capable of binding a peptide fragment of the p53 protein presented in the context of HLA-A2 (page 43 lines 12 to 31). Example 5 and Figure 2 disclose construction of an expression vector encoding a 264 single-chain TCR and IL-2 linked by a peptide linker (page 6 lines 1 to 4, page 35 lines 3 to 21). Examples 7 and 8 and Figure 2 disclose production and purification of soluble single-chain T cell receptor fusion molecules comprising 264 single-chain TCR and IL-2 linked by a peptide linker. Example 9 and Figure 3 disclose that the IL-2 domain of said soluble single-chain T cell receptor fusion molecule has biological activity in stimulating growth of the IL-2 dependent T cell line. Example 10 and Figure 4 disclose that the 264 single-chain TCR domain of said

soluble single-chain T cell receptor fusion molecule has biological activity in binding a peptide (264 peptide) of p53 presented in the context of HLA-A2. Example 11 and Figure 5 disclose that both the 264 single-chain TCR and IL-2 domains of said soluble single-chain T cell receptor fusion molecule have biological activity sufficient to cross-link IL-2 dependent T-cells with antigen presenting cells displaying a peptide (264 peptide) of p53 in the context of HLA-A2. Examples 14 and 15 further disclose use of said soluble single-chain T cell receptor fusion molecules in filling or inhibiting the growth of tumors expressing p53.

The Examiner also believes that there is no support for linkers in single-chain TCR. There is support for $(G_4S)_4$ sequence in single-chain TCRs of examples as noted above and reference to other applications describing linkers in more detail.

Examiner also believes that there is not support for limitation in claim 149. Applicants have amended claim 149 to overcome this rejection.

Based on the foregoing, Applicants believe that there is support in the specification for the claims as pending. Accordingly, Applicants respectfully request reconsideration and withdrawal of the foregoing rejections.

Rejection of Claims Under 35 USC 103

The Examiner rejected the pending claims under 35 USC 103(a) as being unpatentable over Weidanz et al. in view of Bonneville et al. and as being unpatentable over Weidanz et al. in view of Bonneville et al. and further in view of Theobald et al. Applicants traverse this rejection.

The claims as amended are directed to soluble single-chain T cell receptor fusion molecules comprising a T cell receptor and a cytokine or fragment thereof connected by a first peptide linker, wherein the soluble single-chain T cell receptor has one recognition binding site and the cytokine or fragment thereof has a different recognition binding site, wherein the soluble single-chain T cell receptor comprises α and β variable chain TCR covalently linked together by a second peptide linker.

Specifically, Weidanz et al. teach that a soluble single chain TCR comprising an effector molecule linked to the single chain TCR via an Ig-C_L chain. Weidanz et al. teach that the effector molecule can be a cell toxin or biologically active fragment thereof, a chemotherapeutic drug or a detectably-labeled radionuclide molecule suitable for diagnostic or imaging studies (Weidanz page 32 line 32 to page 33 line 19). Weidanz et al. do not teach that the effector

molecule is a cytokine. Additionally, the effector molecules taught by Weidanz et al. are structurally and functionally very different than a cytokine molecule. It is unpredictable whether a cytokine domain will fold correctly as part of a TCR fusion molecule such that it retains receptor finding capability. For example, the effector molecules taught by Weidanz et al. do not recognize receptors on the surface of effector cells in order to mediate their activities.

Weidanz et al. teach a Ig-C_L chain linking the effector molecule and the TCR. The Ig-C_L chain (or functional fragment) between about 70 to 150, preferably between about 90 to 120, and more preferably between about 100 to 110 amino acids in length (Weidanz page 18 lines 23 to 25). In contrast the current application disclose a peptide linker between the single-chain TCR and the cytokine wherein the peptide linker is preferably from about 7 to 20 amino acids, more preferably from about 8 to 16 amino acids (page 19 lines 23-24). Thus the structure of the peptide liker of the current application and the linking Ig-C_L chain of Weidanz are different. (Although not cited by the Examiner, Weidanz does disclose linking the TCR and effector molecule with a second linker.)

Bonneville teaches soluble heterodimeric (two chain) T cell receptors (T receptors) comprising VαCα and VβCβ subunits or other combinations of Vγ-Cγ and Vδ-Cδ subunits (column 2 line 39 to column 3 line 6). Bonneville does not teach a single-chain T cell receptor. Bonneville also teaches a fusion protein between a soluble T receptor and a peptide sequence, the peptide sequence being constitutive of a peptide or of a protein, the fusion protein is obtained by fusing DNA sequence encoding the peptide or protein to one of the chains or to the two chains of DNA encoding the subunits of a T receptor from which their transmembrane portions has been deleted, followed by a co-transfection of the DN sequences thus fused into a host cell (column 3 line 42 to 50). Bonneville teaches that the peptide sequence is IL-2 (column 3 lines 52-53). Thus, Bonneville teaches a soluble heterodimeric (two-chain) TCR directly fused to one or two IL-2 proteins without a peptide liker between the TCR subunits that the IL-2 protein. In contrast the claimed TCR fusion protein comprises a single-chain TCR fused to a cytokine with a peptide linker that allows effective positioning of the biologically active molecule with respect to the TCR molecule binding groove so that the T cell receptor can recognize presenting MHC-peptide complexes and the biologically active molecule can modulate the activity of a cell either to induce or to inhibit T-cell proliferation, or to initiate or inhibit an immune response to a particular site (page 15 lines 6-18). Therefore, the claimed TCR fusion protein and that taught by Bonneville differ in structure in several respects: 1) Bonneville's TCR domain is a two-chain

construct comprising two TCR variable-constant domain chains whereas the TCR domain of the invention is a single chain construct comprising a Vα chain linked to a Vβ chain and 2) Bonneville's IL-2 domain is fused directly to the TCR domain whereas the cytokine domain of the invention is linked to the TCR domain by a linker sequence that allows effective positional of the two domains to permit functional activity.

Bonneville does not specifically exemplify the construction or characterization of soluble TCR proteins comprising a fused IL-2 domain. In addition, Bonneville does not disclose any functional activity of the fused IL-2 domain in the TCR fusion protein. The Examiner states that IL-2 is specific for recognition of effector cells (immune cells expressing IL-2 receptors such a activated T cells). However, Bonneville does not teach that the IL-2 domain of the TCR fusion protein retains this or any of the other known biological activities of IL-2. Given that the IL-2 domain is fused directly to the TCR domains in the constructs of Bonneville, it is uncertain whether the IL-2 domain is capable of binding IL-2 receptor expressed on immune cells due to changes in the fused IL-2 domain structure or steric hindrance by the adjacent TCR domains. As it was known that both the N-terminal and C-terminal domains of IL-2 are important for its biological activity (see for example, Ju et al. 1987. J. Biol. Chem. 262:5723), one skilled in the art would expect that the IL-2 domain of Bonneville directly fused to either the C-terminus or N-terminus of the TCR chain(s) would not retain biological activity. In contrast, the claimed soluble TCR fusion protein comprising a fused biologically active cytokine that is specific for recognition of an effector cells and can modulate the activity of a cell either to induce or to inhibit cell proliferation, or to initiate or inhibit an immune response (claim 86, page 15 lines 6-18, page 19 lines 14-21). Construction, production and characterization of such TCR fusion proteins are shown in Examples 5 - 11, 15 and 16. For example, the biological activity of the fused IL-2 domain of the invention to induce cell proliferation of an IL-2 dependent T cell line is demonstrated in Example 9.

Combining the teachings of Weidanz et al. with Bonneville would not lead to the claimed invention. As indicated, the structure of the TCR fusions of Weidanz et al. and Bonneville are different from each other and from the TCR fusions of the invention. For example, neither Weidanz et al. nor Bonneville disclose a peptide linker between a single-chain T cell receptor and a cytokine that effectively positions these domains such that the T cell receptor can recognize presenting MHC-peptide complexes and the cytokine can recognize immune effector cells. In addition, there is a complete lack of disclosure by both Weidanz et al. and Bonneville

as to the functional activity of the cytokine domain of the TCR fusion molecule that is provided in the claimed invention.

Moreover, the claimed molecules have a number of beneficial characteristics that were unexpected and that are not taught or suggested by the cited references alone or in combination. Submitted herewith is a declaration by Hing Wong, Ph.D., an inventor of the instant application and the President and CEO of Altor Bioscience Corporation detailing the unexpected and surprising results demonstrated by the claimed molecules. For convenience, the contents of the declaration will be summarized below.

The declaration sets forth a number of experiments that show, unexpectedly, that the claimed fusion molecules have highly enhanced efficacy when compared to the any portion of the molecules alone. Specifically, the claimed fusion molecules exhibit longer cell surface residency time and bind more stably to their cell surface receptors than do the cell surface ligands when not part of the fusion molecules. Specifically, the declaration sets forth data demonstrating that the claimed fusion molecules (c264scTCR/IL-2 and MART-1scTCR/IL-2) exhibit longer cell surface residency time and bind more stably to the IL-2 receptor than does IL-2 when not part of the claimed fusion molecules. The experiments further demonstrate that the claimed molecules, (264scTCR/IL-2 and MART-1scTCR/IL-2) showed equivalent IL-2 biologic activity *in vitro* and *in vivo*.

The declaration also details a set of experiments that demonstrate that scTCR/IL-2 fusion proteins unexpectedly have a much longer serum half-live and higher serum recovery than rhIL-2 alone. The longer half-life, modest tissue distribution, slow clearance and stable bifunctionality of the scTCR/IL-2 fusion proteins provide significantly more favorable pharmacokinetic properties than are observed for IL-2-based therapeutic agents.

The experiments described above, and detailed in the accompanying declaration, result in unexpectedly enhanced efficacy of the claimed molecules. For example, the data in the declaration demonstrates that the claimed scTCR/IL-2 fusion proteins have significantly greater efficacy against well-established human xenograft tumors than does rhIL-2 alone. Specifically, treatment with 264scTCR/IL-2 led to marked inhibition of tumor growth and partial to complete regression of tumors in mice by the completion of the dosing regimen, while tumors in mice

administered rhIL-2 alone continued to grow at a rapid rate increasing over 4 fold during the course of treatment.

Accordingly, based on the arguments submitted above, and the experiments detailed in the declaration, the pending claims would not have been obvious to one of skill in the art at the time of filing the instant application. Applicants respectfully request reconsideration and withdrawal of the foregoing rejection.

CONCLUSION

In view of the above amendment, applicant believes the pending application is in condition for allowance.

Dated: March 19, 2008

Respectfully submitted,

By _____

Jonathan M. Sparks, Ph.D.
Registration No.: 53,624
EDWARDS ANGELL PALMER & DODGE LLP
P.O. Box 55874
Boston, Massachusetts 02205
(617) 517-5543
Attorneys/Agents For Applicant